

Inheritance of chloroplasts and mitochondria in Conifers: A review of paternal, maternal, leakage and facultative inheritance

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ABSTRACT

The inheritance of chloroplasts and mitochondria in conifers is reviewed. Generally, in the Pinaceae, chloroplasts are paternally inherited and mitochondria maternally inherited. In contrast, most other conifers have paternal inheritance for both chloroplasts and mitochondria. Several examples of leakage are discussed where normally paternal cp or mt are inherited by 1-3% maternal leakage. A few *Pinus* taxa with mt maternal, have leakage of 1-5% paternal. An unusual case of facultative inheritance is discussed where both forward and reciprocal crosses of *Pinus mugo* x *P. sylvestris* resulted in hybrids with *P. mugo* chloroplasts. Published on-line www.phytologia.org *Phytologia* 101(2):134-138 (March 21, 2019). ISSN 030319430.

KEY WORDS: inheritance, chloroplasts, mitochondria, conifers

It is estimated that in angiosperms, chloroplasts (cp) are maternally inherited in over 70% of the genera (Hipkins et al. 1994) and about 25% of genera are biparentally inherited (Harris and Ingram (1991). Interestingly, chloroplasts were found to be only maternally inherited in dicot trees according to Rojara and Dancik (1992). However, in conifers, chloroplasts and mitochondria (mt) exhibit considerable variation in their inheritance (Table 1).

In the Pinaceae, chloroplasts are generally inherited paternally (via pollen) and mitochondria generally maternally inherited (Table 1). However, maternal and paternal leakage has often been reported (Table 1). Maternal leakage refers to persistence of a few (or sometimes many) chloroplasts and/or mitochondria from the female egg, due to the incomplete degradation during fertilization (Mogensen, 1996). Likewise, paternal leakage is the survival of a few (or many) chloroplasts and/or mitochondria from the pollen during fertilization (Mogensen, 1996). Mogensen (1996, figs. 24-29, etc.), in his seminal paper, gives lucid details for mechanisms for the destruction of cell organelles and he writes several times how “this mechanism could lead to some ‘leakiness’ of maternal plastids”, “resulting in proembryo cells **typically** (emphasis added) containing **only** male cytoplasm”, and “in the Pinaceae, the mechanism of cytoplasmic transmission described above could lead to some leakiness”.

Scmidt et al. (1987) reported leakiness in *Larix decidua* x *leptolepis* in which 5 hybrids exhibited paternal inheritance, but one of the 6 hybrids had maternal chloroplast DNA, and 2 other hybrids had mixed Bam-HI restriction patterns indicative of recombination between paternal and maternal cp DNAs.

Analyses of mitochondria DNA RFLPs of 125 hybrid seedlings originating from 23 controlled crosses of *Pinus banksiana* x *P. contorta* revealed 119 maternal and 6 paternal mitochondria (~5%, Wagner, et al. 1991). Cato and Richardson (1996) utilized cpSSRs to examine inheritance of chloroplast in 206 progeny obtained from controlled pollinations of 4 *Pinus radiata* families. They found 99% of the progeny inherited their chloroplast paternally (pollen). But, approximately 1% of the progeny contained maternal chloroplasts, thus showing leakage in chloroplast inheritance.

Owens and Morris (1991) studied cytoplasmic inheritance in *Pseudotsuga menziesii* and noted that “most of the proembryo cytoplasm is of paternal origin, but some maternal organelles may be included”. They estimated that about 10% of the final number of mitochondria were carried to the proembryo, along with paternal organelles.

Recently, Kormutak et al. (2017, 2018) have reported a most unusual inheritance of chloroplasts in reciprocal crosses of *Pinus mugo* and *P. sylvestris*. They found *P. sylvestris* (female) x *P. mugo* (male) yielded the typical *Pinus* inheritance of paternal (*P. mugo*) chloroplasts in the hybrids. However, *P. sylvestris* (male) x *P. mugo* (female) resulted in all hybrids with *P. mugo* chloroplast DNA (maternally inherited!). If this case can be generalized to natural hybridization of *P. sylvestris* and *P. mugo*, then the analysis of hybrids with only *P. mugo* chloroplasts will likely make studies of hybridization and introgression between these species very difficult. In any case, this example does offer a novel mechanism for chloroplast capture.

The phylogenetic distinctiveness of the Pinaceae is demonstrated by the shift in mode from cp (paternal), mt (maternal) in the Pinaceae to cp (paternal), mt (paternal) in other conifer families (Table 1), although the Taxaceae (*Taxus bacata*) may have cp (paternal), mt (maternal), but the literature is unclear as Mogensen (1996) cites Pennell and Bell (1988) who concluded “There is the **possibility** (emphasis added) that, as in some other gymnosperms, the plastids and mitochondria in the zygote come **in part** from the male gametophyte”.

Cunninghamia konishii (Cupressaceae) seems to be an exception to all conifers (Lu, et al. 2001) in having maternal inherited chloroplasts (Table 1), with the exception of the unusual case of *P. mugo* x *P. sylvestris* (above).

As in the case with the *Pinaceae*, maternal leakage has been reported in other conifer families (Table 1). Ohba et al. (1971) examined the inheritance of the Wogon-Sugi trait (i.e., plants having white-yellowish leaf tips, a chlorophyll mutation) in *Cryptomeria japonica*. They noted “for Wogon-Sugi (,) the trait was **mostly** (emphasis added) transmitted to the progeny when Wogon-Sugi (plant) was used as pollen parent”. So, apparently, some maternal leakage occurs in *Cryptomeria japonica*.

Recently, Kou et al. (2014) re-examined the classical intergeneric hybrid, Leyland cypress (*Chamaecyparis nootkatensis* x *Cupressus macrocarpa*, now *Callitropsis nootkatensis* x *Hesperocyparis macrocarpa*). Keith Rushforth and I provided historical information on the maternal seed sources of the Leyland cypress analyzed in the study (Tables 2, 3, Kou et al. 2014). Six Leyland cypress cultivars, and putative parents or exemplars were analyzed using sequences from 3 mt genes (*coxI*, *atpA*, and *rps3*) and 2 cp genes (*matK* and *rbcL*). Four cultivars from seed of *H. macrocarpa* trees (pollen ex *C. nootkatensis*) had *C. nootkatensis* type chloroplasts and mitochondria. However, two cultivars whose seed purportedly came from a *C. nootkatensis* (maternal) tree (x pollen of *H. macrocarpa*), were grown and contained 2 unusual seedlings: Green Spire and Haggerston Grey. These 2 cultivars were found to have *H. macrocarpa* type chloroplasts and mitochondria. Thus, if the historical records on seed source are correct, the six Leyland cypress cultivars contained 4 showing paternal inheritance and 2 exhibited maternal inheritance. Unfortunately, these results rest on historical records of plant growth and plantings, which are, of course, subject to error.

Another Cupressoideae taxon, *Chamaecyparis obtusa*, was studied (Shiraishi et al. 2001) by use of a cpDNA region, trnD-trnY. In 361 progenies, 352 (97.5%) had the same haplotype as their male parent (paternal) and 9 (2.5%) had the haplotype of their female parent (maternal), showing that although chloroplasts are mostly inherited paternally in *Chamaecyparis obtusa*, there is maternal leakage.

Table 1. Inheritance of cp (chloroplasts) and mt (mitochondria) in conifers. ns = not studied.

	cp	mt	ref.
Pinaceae	pat	mat	review, Mogensen, 1996.
<i>Larix decidua</i> x <i>leptolepis</i>	pat ¹	mat	DeVerno et al. 1991; Schmidt et al. 1987
			¹ 1 hybrid had mat cp (i.e., leakage)
<i>Picea abies</i>	ns	mat	Grivet, et al. 1999
<i>Picea</i> sp.	pat	mat	Sutton et al. 1991
<i>Pinus</i> sp.	pat	mat	Wagner et al. 1987; Neale and Sederoff, 1988, 1989
<i>Pinus banksiana</i> x <i>contorta</i>	ns	mat ²	Wagner et al. 1991
			² ~5% pat leakage
<i>Pinus echinata</i> , <i>Pinus elliotii</i> , <i>Pinus palustris</i> , and <i>Pinus taeda</i>	pat	ns	Wagner et al. 1992
<i>Pinus radiata</i>	pat ³	ns	Cato and Richardson, 1996
			³ ~1% mat leakage
<i>Pinus sylvestris</i> (female) x <i>P. mugo</i> (male)	pat	ns	Kormutak, et al. 2017, 2018
<i>Pinus mugo</i> (female) x <i>P. sylvestris</i> (male)	mat	ns	Kormutak, et al. 2017, 2018
<i>Pinus taeda</i>	pat	mat	Neal and Sederoff, 1988, 1989
<i>Pseudotsuga menzesii</i>	pat ⁴	mat ⁴	Marshall and Neale, 1991; Wagner et al. 1989
			⁴ some organelles from mat(cp) or pat(mt) may be included. Owens and Morris, 1991
Taxaceae			
<i>Taxus bacatta</i>	pat?	mat?	Pennell and Bell, 1988, Mogensen, 1996.
Araucariaceae			
<i>Agathis robusta</i>	pat	pat	Kaur and Bhatnager, 1984
Cephalotaxaceae			
<i>Cephalotaxus drupacea</i>	pat	pat	Gianordoli, 1974; Singh, 1961
Cupressaceae			
Cunninghamioideae			
<i>Cunninghamia konshii</i>	mat	ns	Lu, et al. 2001
Sequoioideae			
<i>Sequoia sempervirens</i>	pat	pat	Neale, Marshall and Sederoff, 1989
Taxodioideae			
<i>Cryptomeria japonica</i>	pat ⁵	ns	Ohba et al. 1971
			⁵ some mat leakage
Callitroideae			
<i>Callitris</i> (4 species)	pat	ns	Sakaguchi, et al. 2014
Cupressoideae			
Leyland cypress - <i>Callitropsis nootkatensis</i> (= <i>Chamaecyparis nootkatensis</i>) x <i>Hesperocyparis</i> (= <i>Cupressus</i>) <i>macrocarpa</i>	4 plants: pat	pat	Kou, et al. 2014
	2 plants: mat	mat	Kou, et al. 2014
<i>Calocedrus decurrens</i>	pat	pat	Neale, Marshall and Harry, 1991
<i>Chamaecyparis obtusa</i>	pat ⁶	ns	Shirashi et al. 2001
			⁶ ~2.5% mat leakage
<i>Chamaecyparis obtusa</i> x <i>pisifera</i>	pat	pat	Kondo, et al., 1998
<i>Chamaecyparis lawsonia</i>	pat	pat	Chesnoy, 1973
<i>Platycladus orientalis</i>	pat	pat	Chesnoy, 1969
<i>Hesperocyparis arizonica</i> x <i>H. macrocarpa</i>	pat	ns	Adams et al. 2018
<i>Juniperus ashei</i> , <i>J. pinchotii</i> , <i>J. virginiana</i>	pollen	pollen	Mohanty et al. 2016, ultrastructural presence of cp and mt in pollen was confirmed by TEM and DNA.

Mohanty et al. (2016) examined the pollen of *Juniperus ashei*, *J. pinchotii* and *J. virginiana* by TEM (Transmission Electron Microscopy) and verified the presence of chloroplasts and mitochondria in pollen in these three species. They also perfected a method to extract DNA from single pollen grains and verified specific cpDNA markers that distinguished these three species. Unfortunately, they did not have access to hybrids from controlled crossings of junipers, so they could not comment on the inheritance of chloroplasts and mitochondria in *Juniperus*.

However, Scion Ltd., New Zealand has made controlled crossings in a closely related genus, *Hesperocyparis*. Adams et al. (2018) recently analyzed 18 hybrids from a single, controlled cross, *H. arizonica* (male) x *H. macrocarpa* (female), and all 18 had perfect *H. arizonica* (paternal) chloroplast DNAs, confirming paternal inheritance of chloroplasts in *Hesperocyparis* (Table 1).

In conclusion, from this brief review of the inheritance of chloroplasts and mitochondria in conifers, it seems very apparent that the literature is divided by the earlier, genetics works (mostly before 1996) and the newer, more molecular reports (1996- present), that will superseded by NextGen technologies, sequencing individual DNA stands, that will very likely reveal unusual modes of variation. We will surely discover more unusual mechanisms of chloroplast and mitochondria inheritance similar to the case of facultative selection reported by Kormutak et al., (2017, 2018).

ACKNOWLEDGEMENTS

This research was supported in part with funds from Baylor University.

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